

(Under this application, which was originally made under Section 91 of the Patents and Designs Acts, 1907 to 1932, a specification was laid open to public inspection on July 1, 1931.

PATENT SPECIFICATION



Application Date: May 24, 1932. No. 14,950/31.

393,319

Complete Accepted: June 8, 1933.

COMPLETE SPECIFICATION.

Medicinal Preparation and Process of Producing same.

(A communication to me from abroad by
McKESSEN & ROBBINS, INCORPORATED,
a Corporation of the State of Connecticut,
United States of America, of
5 Bridgeport, County of Fairfield, and
State of Connecticut, United States of
America.)

I, ALBERT LEVY MOND, Doctor of
Science of the University of Geneva,
10 Chemical Engineer and Chartered Patent
Agent, of 19, Southampton Buildings,
Chancery Lane, London, W.C. 2, a
British Subject, do hereby declare the
nature of this invention and in what
15 manner the same is to be performed, to
be particularly described and ascertained
in and by the following statement:—

The present invention is directed generally to the production of a medicinal preparation for the treatment of anemia and/or pernicious anemia and more particularly to such a preparation which is conducive to the regeneration of hemoglobin in the blood. Hemoglobin contains iron 25 as one of its constituent elements. Efforts have heretofore been made to treat anemia and generally to regenerate hemoglobin by the introduction into the system of iron in various forms. It is the 30 object of the present invention to provide a preparation which greatly enhances the iron metabolism or hematopoiesis, increases the hemoglobin content of the blood and thereby provides effective treatment of anemia and/or pernicious anemia.

The preparation, therefore, contains an iron compound, preferably in the form of a proteid, and more particularly an iron nucleinate or iron peptone, an addition agent which assists in and probably functions as a catalyst in iron metabolism or in the formation of hemoglobin and a fruit acid salt such as sodium citrate. More particularly this addition agent is a 40 suitable copper compound such as a copper proteid, and preferably a copper nucleinate or copper caseinate or a mixture of the same.

The preferred form of the preparation,

therefore, contains iron in one or more of the forms set forth above copper in one or more of the forms set forth above, and a fruit acid salt such as sodium citrate, the incorporation of these ingredients producing a stability in the iron and copper compounds which would not be possessed by a mere mechanical mixture.

50 My foreign correspondents have found each of the following preparations effective in the treatment of anemia and/or pernicious anemia:

1. Solution of copper nucleinate, iron nucleinate and sodium citrate.
2. Solution of copper nucleinate, iron peptone and sodium citrate.
3. Solution of copper caseinate, iron peptone and sodium citrate.

55 Of the above preparations, the form which contains both the iron and the copper in the form of nucleinates is preferred. Nucleoproteins generally and nucleic acid particularly play an important role in body metabolism. The introduction, therefore, of the iron and copper in this form assists greatly in the absorption of these elements into the system and is effective in the formation of the hemoglobin. Moreover, iron and copper compounds generally have astringent and toxic properties whereas masked in the 60 form of organic compounds and particularly in the form of proteids, these undesirable properties are greatly overcome.

70 The following data are the result of experimental test upon anemic rats.

75 For producing anemia in experimental rats my foreign correspondents found it quite advantageous to feed the mothers the U.S.P.X. diet plus warm milk made from whole milk powder, but without vegetables or meat during the period of lactation. At the age of twenty-one days the litters were removed and placed on cow's whole milk diet. When treated in 80 this manner the test animals were observed to develop a marked case of anemia in from three to four weeks as

evidenced by hemoglobin content of the blood and loss of color in eyes, ears, feet and tails. The average hemoglobin content of the animals at the end of this period is about five grams per one hundred c.c. of blood.

At this point of the test the animals are placed in individual cages and fed daily for six days a week one c.c. of a solution of the compounds investigated in about five c.c. of milk. After consuming this test portion each animal was given about fifty c.c. additional milk daily. Hemoglobin determinations were made weekly by means of a Newcomer hemoglobinometer on the sample of blood taken from the tail of the rat in the usual manner. Weekly weighings were made during the

test period.

In table I are given the average results of hemoglobin determinations and weights of three test animals and three controls. The test animals as shown under chart I received 0.25 mg. copper as nucleinate (29.72 per cent. Cu) and 0.50 mg. iron as iron nucleinate (8.89 per cent. Fe). These figures show that in one week's time after the addition of copper and iron, the hemoglobin increased from 6.7 grams per 100 c.c. blood to 14.6 grams.

TABLE I.

Average weight of Hemoglobin Levels of animals receiving 0.25 mg. Copper as Copper Nucleinate (29.72 per cent. Cu) and 0.50 mg. Iron as Iron Nucleinate (8.89 per cent. Fe.)

TEST ANIMALS (3)		CONTROLS (3)		
	Hemoglobin, grams per 100 c.c. blood	Wgt	Hemoglobin, grams per 100 c.c. blood	Wgt
40	At start 6.7	71	6.1	76
	End of 1 wk. 14.6	80	4.9	86
	End of 2 wks. 14.2	94	4.1	96
	End of 3 wks. 15.2	103	3.9	99
	End of 4 wks. 16.3	111	4.1	100

45 Table II contains the average results of hemoglobin determinations and weights of four test rats receiving similar amounts of copper and iron as copper nucleinate (29.72 per cent. Cu) and iron peptone (17.5 per cent. Fe).

TABLE II.

Average weight and Hemoglobin Levels of animals receiving 0.25 mg. Copper as Copper Nucleinate (29.72 per cent. Cu) and 0.50 mg. Iron as Iron Peptone (17.5 per cent. Fe.).

EXPERIMENTAL ANIMALS (4)		
	Hemoglobin, grams per 100 c.c.	Weight, grams
60	At start 7.0	61
	End of 1 wk. 13.4	71
	End of 2 wks. 13.7	81
	End of 3 wks. 14.4	89

65 Under Table III are given the average results of hemoglobin determinations and weights of four test animals receiving similar amounts of copper and iron as copper nucleinate (9.82 per cent. Cu) and iron peptone (17.5 per cent. Fe.).

TABLE III.

Average weight and Hemoglobin Levels of animals receiving 0.25 mg. Copper as Copper Nucleinate (9.82 per cent. Cu) and 0.50 mg. Iron as Iron Peptone (17.5 per cent. Fe.).

EXPERIMENTAL ANIMALS (4)		
	Hemoglobin, grams per 100 c.c.	Weight, grams
80	At start 3.7	80
	End of 2 wks. 13.8	104
	End of 1 wk. 9.9	8.4
	End of 3 wks. 14.3	133

In Tables IV and V are given the average hemoglobin determinations and weights of six test animals receiving 0.25 mgs. of copper as copper caseinate (4.87 per cent. Cu) and 0.50 mg. iron as iron peptone (17.5 per cent. Fe.).

TABLES IV AND V.

Average weight of Hemoglobin Levels of rats receiving 0.25 mg. Copper as Copper Caseinate (4.87 per cent. Cu) and 0.50 mg. Iron as Iron Peptone (17.5 per cent. Fe.).

TABLE IV.		
EXPERIMENTAL ANIMALS (2)		
	Hemoglobin, grams per 100 c.c.	Weight, grams
	At start 3.5	99
	End of 4 days 7.6	100
	End of 11 days 13.1	108
	End of 18 days 14.4	108

TABLE V.		
EXPERIMENTAL ANIMALS (4)		
	Hemoglobin, grams per 100 c.c.	Weight, grams
	At start 6.6	61
	End of 1 wk. 12.1	72
	End of 2 wks. 12.9	78
	End of 3 wks. 14.0	85

PREPARATION OF COPPER NUCLEINATE FROM YEAST.

Copper nucleinate was prepared from dried brewer's yeast by the following method: Two hundred grams of yeast were mixed with 1000 c.c. distilled water and stirred during the addition of 10 grams NaOH in concentrated solution. While cooling with ice, about 0.8 of the

alkali was neutralized with concentrated HCl (20 grams) and the solution finally made acidic to litmus paper with 30 per cent acetic acid. The residue was allowed to settle over night and the liquid portion filtered through a plaited filter paper. Copper sulphate, chemically pure was then added until the liquid contained about 4-1/2 per cent. CuSO₄ and the mixture thoroughly stirred until all copper sulphate was in solution. A light bluish-green flocculent precipitate was immediately formed and was allowed to settle. The supernatant liquid was decanted and the precipitate washed twice with water by decantation. The residue was then filtered by suction on a Büchner funnel and washed thoroughly with distilled water until free of sulphates. The precipitate was then triturated with ninety-five per cent. alcohol in a glass mortar, filtered by suction and after washing twice with alcohol, treated in a similar manner with ether. The precipitate was finally dried on filter paper in the air and was obtained in the form of a light bluish-green amorphous powder. Yield of copper nucleinate was 6.24 grams. On analysis this compound was found to contain 29.72 per cent. of copper and 10.3 per cent. of phosphorus. No sulphate was present when tested in the usual manner.

PREPARATION OF IRON NUCLEINATE FROM YEAST.

Iron nucleinate was prepared from brewers' yeast in somewhat the same manner as the copper compound, ferric chloride, chemically pure, being used in place of copper sulphate. It was necessary, however, to precipitate the iron compound in sixty per cent. alcohol owing to the partial solubility of iron nucleinate in water. In washing the precipitate sixty per cent. and ninety-five per cent. alcohol was used followed by ether as in the case of the copper compound. An amorphous light brown compound was obtained which on analysis contained 8.89 per cent. iron and 4.21 per cent. phosphorus. Yield from two hundred grams yeast was 7.0 grams.

PREPARATION OF COPPER NUCLEINATE FROM NUCLEIC ACID.

Five grams of nucleic acid was added to 300 c.c. of distilled water in an 800 c.c. beaker and NaOH solution added in small amounts with constant stirring until the nucleic acid went into solution. The solution was then slightly acidified with thirty per cent. acetic acid and copper sulphate, chemically pure, solution added with stirring until precipitation was complete. A light green flocculent precipitate was immediately formed. On settling the supernatant liquid was decanted.

The residue was washed twice by decantation, filtered by suction and washed free of sulphates. It was then triturated with ninety-five per cent. alcohol in a glass mortar, filtered and washed with alcohol. This treatment was then repeated with ether. A light green amorphous powder was obtained containing 9.82 per cent. of copper and 7.05 per cent. of phosphorus. Yield 5.08 grams.

PREPARATION OF COPPER CASEINATE FROM CASEIN.

One hundred grams of purified casein was added to 1500 c.c. distilled water and, while stirring, about 119 c.c. of normal NaOH added in portions until the casein was completely dissolved. Copper sulphate twenty-one grams in 120 c.c. water, was then added while stirring until precipitation was complete. The copper caseinate which separated as a green flocculent precipitate was then filtered by suction and washed with water until free of sulphates. After washing with fifty per cent. alcohol, ninety-five per cent. alcohol and ether, the residue was dried on filter paper in the air. A bluish-green crystalline powder was obtained containing 4.87 per cent. of copper. Yield 101 grams.

The iron peptonate used in these tests was N.F.V. (National Formulary Fifth Edition) powder and contained 17.5 per cent of iron.

It will be seen from the foregoing that 100 solutions of the iron and copper compounds with a stabilising addition, i.e., a fruit acid salt such as sodium citrate constitute an effective treatment of all cases of anemia. These preparations can be 105 made up in liquid form which are exceedingly palatable and the required daily dosage is small. The cost per daily dose is comparatively low. A positive reaction in the regeneration of hemoglobin is 110 obtained.

The following tabulations give the specific compositions of the solutions used in the experiments that are tabulated hereinabove:

SOLUTION OF COPPER NUCLEINATE AND IRON NUCLEINATE.

Copper nucleinate (made from brewers' yeast, 29.72% copper).

Iron nucleinate (made from brewers' yeast 8.89% iron).

Solution made up so that 1 c.c. contains:

0.25 mg. copper or 0.842 mg. copper nucleinate (29.72% copper)	125
0.50 mg. iron or 5.6 mg. iron nucleinate (8.89% iron)	
8.00 mg. sodium citrate (U.S.P. IX [United States Pharmacopeia. Ninth Edition])	130

BEST AVAILABLE COPY

	Formula Used (500 c.c. Solution)	0.25 mg. copper or 5.13 mg. copper caseinate (4.87% copper)	80
	0.421 grams copper nucleinate	0.50 mg. iron or 2.85 mg. iron peptonate (17.5% iron)	
	2.800 grams iron nucleinate	8.00 mg. sodium citrate (U.S.P. IX)	70
	4.000 grams sodium citrate	Formula Used (500 c.c. Solution)	
5	72.00 c.c. alcohol	2.565 grams copper caseinate	
	24.00 c.c. sugar syrup (85 : 100 H ₂ O)	1.425 grams iron peptonate	
	24.00 c.c. glycerin	4.000 grams sodium citrate	
	0.08 c.c. oil of orange (Sweet-Italian)	72.00 c.c. alcohol	75
10	0.08 c.c. acetic ether (ethyl acetate, anhyd.)	24.00 c.c. sugar syrup (85 : 100 H ₂ O)	
	0.01 grams vanillin	24.00 c.c. glycerin	
	Distilled water to make 500 c.c. solution.	0.8 c.c. oil of orange (Sweet-Italian)	
	NOTE.	0.08 c.c. acetic ether (ethyl acetate, anhyd.)	
15	Four (4) teaspoonsfuls (4 c.c. each) of the above copper-iron solution should contain 4.0 mgs. copper and 8.0 mgs. iron.	0.01 grams vanillin	80
	One ounce of this solution contains:	Distilled water to make 500 c.c. solution.	
	0.026 grams copper nucleinate	NOTE.	
20	0.168 grams iron nucleinate	Four (4) teaspoonsfuls of the above copper-iron solution should contain 4.0 mgs. copper and 8.0 mgs. iron.	85
	One ounce contains about 30 c.c. or 7-1/2 teaspoonsfuls.	One ounce of this solution contains:	
	SOLUTION OF COPPER NUCLEINATE AND IRON PEPTONATE.	0.154 grams copper caseinate	
25	Copper nucleinate (made from brewers' yeast 29.72% copper).	0.085 grams iron peptonate	90
	Iron peptonate (N.F.V.-Powder, 17.5% iron).	One ounce contains about 30 c.c. or 7-1/2 teaspoonsfuls.	
	Solution made up so that 1 c.c. contains:	Similar preparations for the treatment of humans preferably contain different proportions of the several ingredients. The following tabulations show the preferred compositions. It will be understood, however, that these compositions are submitted herewith only for illustrative purposes and that the specific proportions of the several ingredients may be varied widely, and some of these may be entirely omitted. The essential feature of these preparations is that they each contain iron and copper in suitable forms and a fruit acid salt, the iron serving to enter into the bodily metabolism for the regeneration of hemoglobin, the copper having apparently only a catalytic function, whilst the fruit acid salt contributes to the stabilisation of the preparations.	95
30	0.25 mg. copper or 0.842 mg. copper nucleinate (29.72% copper)	100	
	0.50 mg. iron or 2.85 mg. iron peptonate (17.5% iron)		
35	8.00 mg. sodium citrate (U.S.P. IX).		
	Formula Used (500 c.c. Solution)		
	0.421 grams copper nucleinate		
	1.425 grams iron peptonate		
	4.000 grams sodium citrate		
40	72.00 c.c. alcohol		
	24.00 c.c. sugar syrup (85 : 100 H ₂ O)		
	24.00 c.c. glycerin		
	0.08 c.c. oil of orange (Sweet-Italian)		
	0.08 c.c. acetic ether (ethyl acetate, anhyd.)		
45	0.01 grams vanillin		
	Distilled water to make 500 c.c. solution.		
	NOTE.		
50	Four (4) teaspoonsfuls of the above copper-iron solution should contain 4.0 mgs. copper and 8.0 mgs. iron.		
	One ounce of this solution contains:		
	0.025 grams copper nucleinate		
55	0.085 grams iron peptonate		
	One ounce contains about 30 c.c. or 7-1/2 teaspoonsfuls.		
	SOLUTION OF COPPER CASEINATE AND IRON PEPTONATE.		
60	Copper caseinate (made from purified casein 4.87% copper)		
	Iron peptonate (N.F.V.-Powder 17.5% iron).		
	Solution made up so that 1 c.c. contains:		
65	0.421 grams copper nucleinate		
	11.424 grams iron nucleinate (8.89% iron)	115	
	9.000 grams sodium citrate		
	150.000 c.c. alcohol	120	
	42.500 grams sugar		
	50.000 c.c. glycerin		
	0.160 grams oil of orange (Sweet-Italian)		
	0.160 grams acetic ether (ethyl acetate, U.S.P.)	125	
	0.020 grams vanillin		
	1000.000 c.c. distilled water to make		
	14.25% (theoretical) alcoholic strength above solution	130	

Maximum dose: Metric, 8 c.c.—
Apothecaries, 2 fluidrachms.
One maximum dose contains about:
0.001 grams copper or 0.0033 grams
5 copper nucleinate.
0.008 grams iron or 0.0913 grams iron
nucleinate.

Daily dosage recommended:
From 1 to 2 teaspoonfuls, in a wine-
10 glassful of milk three times daily just
before meals.

PROCEDURE FOR MAKING SOLUTION.

1. Dissolve the copper nucleinate and
sodium citrate in 500 c.c. distilled water
15 by vigorous stirring. When copper is in
solution add the iron nucleinate and stir
until dissolved.

2. Dissolve the vanillin, oil of orange
and acetic ether in the alcohol and add
20 this solution to the first with stirring.

3. Add the glycerin and sugar and stir
until dissolved.

4. Make up to the proper volume with
distilled water.

25 SOLUTION OF COPPER NUCLEINATE AND
IRON PEPTONATE.

Formula (1000 c.c. solution).

Copper nucleinate
(29.72% copper) - 0.421 grams
30 Iron peptonate
(17.5% iron) - 5.700 „
Sodium citrate - 9.000 „
Alcohol - 150.000 c.c.
Sugar - 42,500 grams
35 Glycerin - 50.000 c.c.
Oil of orange
(Sweet-Italian) - 0.160 „
Acetic ether
(ethyl acetate, U.S.P.) 0.160 „
40 Vanillin - 0.020 grams
Distilled water to make 1000.000 c.c.
Alcoholic strength above
solution, 14.25% (theoretical)

Maximum dose: Metric, 8 c.c.—

45 Apothecaries, 2 fluidrachms.
One maximum dose contains about:
0.001 grams copper or 0.0033 grams
copper nucleinate.

0.008 grams iron or 0.0456 grams iron
50 peptonate.

Daily dosage recommended:
From 1 to 2 teaspoonfuls, in a wine-
glassful of milk three times daily just
before meals.

55 PROCEDURE FOR MAKING SOLUTION.

1. Dissolve the copper nucleinate and
sodium citrate in 500 c.c. distilled water
by vigorous stirring. When copper is in
solution add the iron peptonate and stir
60 until dissolved.

2. Dissolve the vanillin, oil of orange
and acetic ether in the alcohol and add
this solution to the first with stirring.

3. Add the glycerin and sugar and stir
65 until dissolved.

4. Make up to proper volume with dis-
tilled water.

SOLUTION OF COPPER CASEINATE AND IRON
PEPTONATE.

Formula (1000 c.c. solution) 70

Maximum dose: Metric, 8 c.c.—
Copper caseinate
(4.87% copper) - 2.565 grams
Iron peptonate
(17.5% iron) - 5.700 „ 75
Sodium citrate - 9.000 „
Alcohol - 150.000 c.c.
Sugar - 42,500 grams
Glycerin - 50.000 c.c.
Oil of orange
(Sweet-Italian)- 0.160 „
Acetic ether
(ethyl acetate, U.S.P.) 0.160 „
Vanillin - 0.020 grams
Distilled water to make 1000.000 c.c. 85
Alcoholic strength above
solution, 14.25% (theoretical)

Maximum dose: Metric, 8 c.c.—
Apothecaries, 2 fluidrachms.

One maximum dose contains about: 90
0.001 grams copper or 0.0205 grams
copper caseinate.
0.008 grams iron or 0.0456 grams iron
peptonate.

Daily dosage recommended: 95
From 1 to 2 teaspoonfuls, in a wine-
glassful of milk three times daily just
before meals.

PROCEDURE FOR MAKING SOLUTION.

1. Dissolve the copper caseinate and 100
sodium citrate in 500 c.c. distilled water
by vigorous stirring. When copper is in
solution add the iron peptonate and stir
until dissolved.

2. Dissolve the vanillin, oil of orange 105
and acetic ether in the alcohol and add
this solution to the first with stirring.

3. Add the glycerin and sugar and stir
until dissolved.

4. Make up to the proper volume with 110
distilled water.

Having now particularly described and
ascertained the nature of my said inven-
tion and in what manner the same is
to be performed, I declare that what I 115
claim is:—

1) A process for the production of
stabilised solutions of iron-copper pro-
tein compounds, such as, for example,
iron nucleinate or iron peptonate and 120
copper nucleinate or copper caseinate, in
which the protein compounds are
stabilised with the aid of water soluble
fruit acid salts, such as sodium citrate.

2) The process of producing a medicinal 125
preparation for aiding in the regenera-
tion of haemoglobin, substantially as
described.

3) A medicinal preparation for aiding
in the regeneration of haemoglobin, when 130

BEST AVAILABLE COPY

produced according to the process set
forth in the preceding claims.
Dated this 21st day of May, 1931.

ALBERT L. MOND,
19, Southampton Buildings,
Chancery Lane, London, W.C. 2.

Redhill: Printed for His Majesty's Stationery Office, by Love & Malcolmson, Ltd.—1938.